IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant: Kristian Lund Henriksen et al

Serial No. 10/584,113

Filed: 04/04/2007

For PROBIOTIC TABLET FORMULATIONS

Examiner: Laura J Schuberg

Art Unit 1657

Attorney Docket BECK 001

DECLARATION OF MARIANNE WINNING UNDER 37 CFR §1.132

I, Marianne Winning of Rypevænget 108, 2980 Kokkedal, Denmark declare and state

1. Details of my experience in the field to which the Application relates are set out in my C.V. which is annexed hereto. I am one of the inventors of the subject matter of the Application. I consider myself to be, in a minimum, one of ordinary skill the art of the subject matter claimed in this application.

2. I am familiar with the contents of the specification of the Application and I have studied the objections raised or maintained in the Office Action of 13th October 2010. I have studied the prior art references relied upon therein and referred to as Glagau et al, Bakulesh et al and Runge et al.

3. As described in the introductory portion of the specification of the Application, it was known prior to the making of the invention that probiotic micro-organisms can be quite stable on storage if formulated by themselves in dry form with an inert carrier material in the form of a tablet.

4. Formulations of this kind are described in Runge et al at column 19-20. These formulations are based on mixtures of a powder concentrate of micro-organisms and formulation ingredients. The production of the powder concentrates is described earlier at columns 15-18. Example S2 at column 16 is typical. A cell suspension of micro-organism (*Lactobacillus plantarum*) was supplemented with ingredients of a kind which would be beneficial rather than deleterious to the storage and survival of the micro-organism and the mixture was spray dried to form a powder concentrate.

This powder concentrate was used in for instance Example F1 by mixing the powder concentrate with further listed ingredients and the resulting powder mixture was compacted and granulated.

5. Runge et al describes conditions of dryness (water content) and of water activity which are said to be appropriate to maintaining stability of such micro-organism formulations on storage and indicates that the water content should be 'extremely low' and from about 2 to 3%; whilst the water activity should be from 0.03 to 0.15.

6. The formulations of Runge et al are not disclosed as containing any of iron, copper, vitamin B6, zinc, manganese, chromium, pantothenic acid and pantothenic acid salts. Such ingredients are not included in starter cultures and would have no useful role in such starter cultures. Starter cultures are intended to be mixed with large amounts of liquid and if any of these aggressive ingredients were desired to be present in use, one would supply them with the liquid component and avoid entirely the difficulty of exposing the micro-organism to them in the dry state. Accordingly, the conditions described in Runge et al are not designed to be suitable for the inclusion of these aggressive ingredients in the dry formulations.

7. Prior to the making of the invention, it was believed that if these materials were included in a composition of the kind described by Runge et al, the viability of the micro-organism would be substantially damaged. Such a belief is reflected in US6254886 (Fusca et al). Thus, at column 1, lines 42-50, Fusca et al taught that formulations of micro-organisms containing any of minerals, fats, vitamins, carbohydrates, proteins, bulk materials, or trace elements are extremely unstable with respect to the viability of the micro-organisms. They aimed to produce a formulation containing not only probiotic micro-organisms but also "vitamins, minerals, bulk materials etc." which remained stable despite these additives (column 4, lines 59-62). Their solution was first to separate the micro-organisms into a separate layer of a multilayer tablet, putting the additives into their own separate layer and secondly to dry the ingredients to an extreme level, much drier than suggested in Runge et al. They stipulated a water content of not more than 0.1% for the layer containing the micro-organisms as being a **necessity** (column 2, lines 33-36). This is a level of dryness 20 times greater than in Runge et al (0.1% vs 2% or more).

8. Fusca et al also reported a surprising finding that such a multilayer formulation produced a level of stability of the micro-organisms which was actually greater than if the additive containing layer was absent (as it would be in Runge et al).

9. Runge et al of course contains no teaching regarding multi-layer tablets or regarding any formulations which are to include any of iron, copper, vitamin B6, zinc, manganese, chromium, pantothenic acid and pantothenic acid salts. I believe that the reader of Runge et al, aware of Fusca et al, would have had a strong expectation that the levels of water and water activity taught in Runge et al would only be applicable

to achieve good viability if these materials were excluded given the explicit teaching in Fusca et al that a water content of no more than 0.1% is essential, a necessity.

10. When I and my co-workers made the invention as claimed we were accordingly very surprised that it was found possible to achieve a good level of viability on storage in multi-layer tablets having a much higher water level than is specified in Fusca et al, even when the tablets contained some or all of the deleterious materials discussed above. It was our expectation prior to making the invention that the relatively high water content of the tablets according to the invention would have facilitated the action of these materials in adversely affecting viability.

11. Glagau et al describes a solution to the problem of providing both probiotic micro-organisms and other nutritionally active ingredients without the viability of the micro-organisms being adversely affected. Their solution is to provide two separate components. This is exemplified for instance in Example 1 as being in the form of two separate sets of granules I and II, where I contains probiotic micro-organisms and II contains nutritionally active materials including iron, vitamin B6 and pantothenic acid. There is no disclosure of water content or activity but since the probiotic micro-organisms are present only in a separate component, no special water content or activity would be required in that component for stability, provided that it is reasonably dry. For instance, that component could have the levels of water content and water activity taught by Runge et al. It would not need the extreme drying taught in Fusca et al. It is clear to me from reading Glagau et al as a whole that they do not intend the 'separate' components to be joined in a single unit as in a multi-part tablet but intend them to be fully physically separate.

12. A combination product of that kind however in which there are two separate sets of granules would not be in accordance with our invention as claimed, whatever water content and water activity might be used.

13. A reading of Glagau et al would not have made it obvious to me that a multi-layer tablet could contain both probiotic micro-organisms and at least one of iron, copper, vitamin B6, zinc, manganese, chromium, pantothenic acid and pantothenic acid salts and have good stability without extreme drying. On the contrary, the splitting of the combination product in Glagau et al into two components so as to keep the probiotic micro-organisms separate from the other ingredients would have encouraged my belief that a multi-layer tablet would not be a viable proposition without the extreme drying taught by Fusca et al.

14. I would not have seen anything in Runge et al to change this perception because Runge et al teaches conditions that are described only to keep the viability of micro-organisms which do not face the challenge of having in the same tablet any of iron, copper, vitamin B6, zinc, manganese, chromium, pantothenic acid and pantothenic acid salts. It would not have seemed to me that Runge et al was suggesting conditions that could be expected to work in a multi-layer tablet containing at least one of iron,

copper, vitamin B6, zinc, manganese, chromium, pantothenic acid and pantothenic acid salts, given the teaching of Fusca et al. I believe that the expectations of those of ordinary skill in the art generally upon reading these disclosures would have been similar to mine.

15. Bakulesh et al describes tablets in which an anti-infective agent and a probiotic micro-organism coexist separated by a water barrier. This barrier may take the form of a coating applied to one or the other of the anti-infective agent and the probiotic. No water content or activity is discussed. It is suggested by the Examiner that it would have been obvious following a reading of Glagau et al and of Bakulesh et al that one could make a multi-zoned single tablet as the product of Glagau et al using barrier materials as per Bakulesh et al to keep the deleterious ingredients away from the probiotic micro-organisms and that if one did this it would be obvious to use the water content and water activity taught by Runge et al.

16. In my view the person of ordinary skill in the art, nor myself, would not have found it obvious to combine the teachings of Glagau et al and Bakulesh et al as alleged by the Examiner for several reasons. First, the clear disclosure in Glagau et al is for multiple physically completely separate components, not for reducing the multiple components into a single unit such as one tablet. Thus, Glagau et al teaches at the last paragraph of page 4 of the translation that the "at least two separate" products involved should preferably actually be three or even better four separate components and also teaches that having separate components enables separate administration of each component. Thus, moving to one combined multi-layer tablet is contrary to the teaching expressed in Glagau et al. I would not have found it obvious based on Glagau et al and Bakulesh et al to produce a Glagau et al type of product in the form of a multi-zone tablet.

17. Secondly, Glagau et al and Bakulesh et al are not concerned with the same type of ingredients which are deleterious to the micro-organisms. In Glagau et al, as in the present invention, these include materials such as iron, zinc, manganese and copper which are very small ionic species that are expected to be very mobile within a formulation. In Bakulesh et al, the problem ingredients are anti-infective agents exemplified by ampicillin. These are much larger molecular species which I would expect to be much easier to retain in a separate tablet compartment. I would not have expected it to be possible to prevent migration of metal ions between zones by the use of a water barrier over the necessary storage lifetime of a probiotic tablet. Over a prolonged storage period, such ionic species would be expected to migrate along with water even through this kind of barrier coating. Moreover, metal ions of this kind are very aggressive in their action which is catalytic such that the same metal ions are able to reduce the viability of many organisms. An anti-infective molecule will typically not be able to kill a large number of organisms but will be consumed in carrying out its action. So I would have found no assurance from Bakulesh et al that it would be possible compartmentalise materials such as iron, zinc, manganese and copper within a multi-layer tablet such that it would not be necessary to adopt the

extreme drying taught by Fusca et al.

18. Moreover, Bakulesh et al does not describe the extent of drying to be employed and is not inconsistent with Fusca et al in this respect. Even recognising that anti-infective agents are likely to be easier to contain and less aggressive than metal ions, I would have considered it likely that if the method of Bakulesh et al was to work even with anti-infectives, it would have been an unstated but necessary aspect of the method that one would use extreme drying as taught by Fusca et al. Our finding that such extreme dryness is in fact not needed even to prevent metal ions migrating and damaging viability provided that the water activity is sufficiently low remains in my view surprising following a reading of all of the cited art.

19. Moreover, I note that the teaching of Bakulesh et al is not backed up by any real data showing that the formulations taught are genuinely viable after prolonged storage as alleged. I believe that the skilled worker in this field would doubt very much that the teaching of Bakulesh et al is anything more than wishful thinking and speculation. Some of the storage times quoted are clearly just fanciful, for instance 36 months for a liquid suspension in Example III. This robs all of the storage times given of credibility. However, even if I suppose that some of the storage results in Bakulesh et al are valid and even if I additionally suppose that anti-infectives can be tolerated if one applies the water content and water activities of Runge et al to formulations as in Bakulesh, it still does not in my view follow that from these teachings one could deduce that one could obtain stability on storage if instead of anti-infective agents one had present the aggressive materials with which the invention is concerned, given their different level of aggression and their greater mobility compared to anti-infectives. From the teachings of Glagau et al regarding separation and the teachings of Fosca et al regarding drying, I believe that the reader of ordinary skill in the art taking all of these teachings into account would not have expected to obtain viability using the conditions of water content and activity characterising the invention when nutritionally active ingredients are present as claimed.

20. I note the Examiner's statement at page 14 of the current Office Action that 'the teaching of Runge et al is applicable to any formulation that contains a probiotic and wishes to maintain the viability of that probiotic in the formulation'. I do not agree with that statement. I see no explicit assertion to that effect in Runge et al. Runge et al. is concerned only with starter cultures for industrial processes, not with multi-layer tablet formulations for human consumption. The problems associated with such formulations will have been of no concern to Runge et al. The water content and activity recommendations of Runge et al were made only in the context of starter cultures free of course from the deleterious ingredients that are included in the tablets of the invention and in the multi-component products of Glagau et al. Runge et al simply teaches me nothing as a skilled person regarding the water content and water activity appropriate for multi-zone tablets of the kind claimed in the application.

21. I note that in the current Office Action, the Examiner states that Glagau et al teaches that the product may be formulated as a multi-component single tablet at page 5, 4th paragraph. As an experienced worker in this field, I see no such teaching there. I would agree that Glagau et al teaches that there may be one tablet, but I see no teaching that this one tablet may be a multi-component tablet containing both the probiotic and the nutritional ingredients such as iron, zinc, manganese and copper. My understanding of this passage is that the product may consist of tablets, capsules, solutions and/or granulates. There have to be at least two separate components, one for the probiotic and one for the other ingredients, although preferably as discussed above there are more than two. One can choose freely for each of the two components as to whether it is a tablet, capsule, solution or granulate. Hence there may be zero tablets involved and equally there may be one tablet involved. But that would imply only that where there are zero tablets there must be at least two of capsules, solutions or granulates and that where there is one tablet involved there must be at least one solution, capsule or granulate. Only this way does one satisfy both Glagau et al's requirement for separating the probiotic from the other ingredients whilst at the same time having one tablet. Contrary to the Examiner's assertion nowhere in Glagau et al in my view is there a teaching of a multi-component single item, whether tablet, capsule, solution or granulate, that contains in the one item both the probiotic and the other ingredients. Such an item indeed is entirely contrary to Glagau et al's teaching a whole and is not implied by the reference to the combination product comprising one tablet. It is not said that the combination product consists of one tablet.

22. I further declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true, and that these statements are made with the knowledge that wilful false statements and the like so made are punishable by fine or imprisonment, or both, under §1001 of Title 18 of the United States Code, and may jeopardise the validity of the application or any patent issuing thereon.

Marianne Winning — Date: 15 March 2011

Marianne Winning
Cand.Pharm.

Date: 10 February 2011

Curriculum Vitae

Personal data

Navn:	Marianne Winning
Adresse:	Rypevænget 108, 2980 Kokkedal
Telefon:	+45 4918 0408, mobil +45 4083 7032
E-mail:	marianne.winning@mail.dk
Date of birth	22 May 1947

Summary

Experience in product development and new business development within pharmaceutical, health food and food industries. A combination of technical, scientific and market oriented disciplines has resulted in a range of successful products and new business areas

Experience

2009 – 2010	Free-lance consultant: Ad hoc tasks for the pharmaceutical industry
2001 – 2009	Ferrosan A/S, Health and Life Style Division, Project Manager and New Opportunity Manager: Carried out projects within vitamins, botanical extracts and probiotics. Initiated a production plant for probiotic consumer products built on new and advanced principles. Created a range of product concepts, which has been succesfull on the market. One patent (tableting principle)
1999 – 2001	Chr. Hansen A/S, Bio Ingredients. New Business Development: Built a range of health food ingredients, based on natural food colors.
1991 – 1999	Chr. Hansen A/S, Food Ingredient Division. Marketing Manager of natural colors for food: Developed the marketing function from 4 to 20 employees. The department included application technology, sales support and education of sales staff. The department became ISO certified. 2 patents (natural color formulations and applications).
1974 – 1991	DanoChemo A/S (now BASF Health and Nutrition). Development chemist, Manager of Application Technology, Manager of Product Development, New Business: Developed encapsulation processes and a range of micro encapsulated and film coated products, among others fish oil, vitamin C, flavoring ingredients and natural colors. Promoted the new products and concepts in the food, feed and pharma industries. 3 patents (microencapsulation and applications).
1971 – 1974	Ferrosan A/S. Chemist in Production and Development Lab: Among other things developed first generation of chewable multi vitamin and mineral tablets and a new generation of medical chewing gum. Solved various production problems and optimized processes.

Education

1971	Candidate from Pharmaceutical University Copenhagen
1988	Commercial education from Elsinore Business School
1971-2008	Technical education in microencapsulation, film coating, tablet technology, pharmaceutical formulations, food and feed technologies, drying, grinding and micronisation, mixing, particle technology, water activity, organoleptic and sensoric disciplines, surface chemistry, suspension technology, controlled release, pre-and probiotics, and other topics.
1971-2009	Personal development and training in management disciplines at various levels and in innovation.

Publications and patents

Poster: Microencapsulated Fish Oil, Characteristics and Application. American Oil Chemist Society Annual meeting. New Orleans 1987.

Presentation: Fish Oils in the Dairy Industry. Food Ingredients Europe. London 1988.

Poster: DHA enriched Infant Formula. Internationales Symposium über n-3 Fettsäuren. East Berlin 1988.

Publication: Skelbæk T., Andersen N.G., Winning M., Westergård S.: Stability in fish feed and bioavailability to rainbow trout of two ascorbic acid forms. Aquaculture 1989.

Presentation: Fish Oils in the Dairy Industry. Congrès Internationale Chevreul. Angers, 1989.

Presentation: Application of Dry n-3 in the Food Industry. Lipid Forum, 15th Scandinavian Symposium on Lipids. Rebild 1989.

Publication: Colorants "Micro-cap" una innovación en colorantes naturales. ILE 1992.

Presentation: Natural Colors – Aspects for the Extrusion industry. Internationalt ZDF seminar Tyskland 1994.

Publication: Micro-encapsulated colours – Natural colours with improved stability. Agro-Food-Industry 1995.

Publication: Susan Lawlor, Marianne Winning: Fill the Nutrient Gap with Functional Extracts from Fruit and Vegetables. Nutraceuticals Now, 2000.

Publication: "Chr. Hansen aims at functional food and health food". Plus Proces 2000.

Advitorials, small ad hoc publications and contact to the public press.

Patents

PCT Publication Nr. WO 91/00692
PTC Publication Nr. WO 91/06292
PTC Publication Nr. WO 97/26802
PCT Publication Nr. WO 97/26803
US Patent No. 5460823
US Patent No. 6190686